NEW INSIGHTS INTO THE PATHOGENESIS AND TREATMENT OF CANINE THYROID CARCINOMA M Campos[1], R Ducatelle[1], M Kool[2], J Mol[2], G Rutteman[2], H Kooistra[2], L Duchateau[1], K Peremans[1], I Van de Maele[1], H de Rooster[1], I Polis[1], S Daminet[1]. 1. Ghent University Faculty of Veterinary Medicine, Merelbeke, Belgium. 2. Utrecht University Faculty of Veterinary Medicine, Utrecht, The Netherlands.

The goals of this research were to investigate the presence of mutations and relative expression of candidate genes, to explore new therapeutic targets and to investigate clinical, pathological and immunohistochemical prognostic markers in canine thyroid cancer.

The medical records of Ghent and Utrecht Universities were reviewed (1986-2012). Dogs diagnosed with thyroid neoplasia were included if formalin-fixed paraffin-embedded tumor samples were available.

Each tumor section (n=74) was reviewed and scored by a board-certified pathologist (histologic type, % necrosis, % hemorrhage, nuclear pleomorphism, mitotic index, evidence of vascular or capsular invasion) and immunohistochemistry was performed for calcitonin, Ki-67, E-cadherin, cox-2, VEGF, P-gp and p53. Differentiation between follicular-cell tumors and medullary tumors was based on immunohistochemistry for calcitonin. RNA was isolated from frozen tumor samples (n=62) and frozen healthy thyroid (n=10) and sequencing was performed for specific coding regions of RAS (K, H and N), PIK3CA, PTEN, RET and BRAF genes. qPCR was performed for VEGFR1, VEGFR2, EGFR, PIK3CA, PIK3CB, PTEN, PDPK1, Akt1, Akt2, calcitonin and reference genes HPRT and RPS5.

All tumors were carcinomas. In 1 follicular-thyroid carcinoma (FTC) and 1 medullary thyroid carcinoma (MTC), 2 missense mutations (G12R and E63K respectively) were found in K-RAS.

Relative expression of VEGFR1, EGFR, Akt1, Akt2, and PIK3CB genes was significantly higher in FTC and MTC than in healthy controls, suggesting PI3K/Akt pathway activation.

80% percent of FTC and all MTC exhibited a very high percentage (76-100%) of neoplastic cells immunopositive for VEGF. 13% percent of FTC and 50% of MTC presented cox-2 expression. 7% percent of FTC and 70% of MTC presented P-gp expression. No tumor was immunopositive for p53 expression. VEGF is a potential therapeutic target in both FTC and MTC, while cox-2 and P-gp seem to be attractive molecular targets in canine MTC.

46 dogs (30 FTC and 16 MTC; stage I-III) underwent thyroidectomy and were included in a survival analysis. Outcome was comparable between FTC and MTC. Inappetence (P=0.007), histologic vascular invasion (P=0.027) and surgical vascular invasion (P=0.005) were associated with shorter disease-free survival. Histologic vascular invasion (P=0.009), large tumor diameter (P=0.004) and high Ki-67 labeling index (P=0.004) were associated with shorter time to metastases. Delayed time to presentation (P=0.041) was associated with shorter time to recurrence. E-cadherin expression was not associated with outcome. Intensive monitoring and adjunctive therapy might be indicated when negative prognostic factors are identified.